The use of customized mouthguards during the training produced protective effects on salivary factors of young athletes

A. Chiavaroli*¥, S. D'Ercole**¥, D. Fulco**, C. Ferrante*, G. Orlando*, L. Recinella*, D. Tripodi**

**Department of Pharmacy, University "G. d'Annunzio" of Chieti-Pescara, Chieti, Italy*
***Department of Medical, Oral and Biotechnological Sciences, University "G. d'Annunzio" of Chieti-Pescara,, Chieti, Italy*
¥These authors contributed equally to this work.

e-mail: simonettadercole@unich.it

DOI 10.23804/ejpd.2021.22.03.8

Abstract

Aim Custom-made mouthguards have many advantages compared to the stock and ready-made types, but sport treatments with custom made mouthguards involve changes in ecological factors of the oral cavity. In the present study we investigated the potential protective role of salivary factors, such as pH value, volume, prostaglandin E2 (PGE2) and 8-iso-prostaglandin F2α (8-iso-PGF2α) levels during training with customised mouthguards.

Materials and methods A total of 80 subjects were selected: 40 athletes, of whom 20 practice volleyball and 20 basketball (test group), and 40 subjects who attend a gym at a non-competitive level (control group). The athletes (test group) were analyzed at baseline (T0), pre-training (T1), post-training with custom-made Ethylene-Vinyl-Acetate (EVA) mouthguards (T2), post-training without mouthguards (T3). The control group was analyzed only at baseline (T0). On each player, in the 4 time points, and on the control group at T0, we stimulated saliva for determining PGE2 and 8-iso-PGF2α levels by radioimmunoassay and pH value by a pH meter and volume/ml. Saliva pH was calculated with a pH meter.

Results We observed an inhibition of 8-iso-PGF2α salivary release induced by physical exercise and by use of custom-made mouthguard, while we found an increase in PGE2 salivary level in athletes after training and wearing the mouthguard. Furthermore, in the test of the volume of saliva produced in 5 minutes, a significant inhibition of saliva production emerged in the athletes who did not use the mouthguard during sports activities.

Conclusion Sports activity could lead to a reduction in oxidative stress and the use of mouth guards seems even more effective for athletes.

KEYWORDS Custom-made mouthguard, Training, 8-iso-PGF2α, PGE2.

Introduction

Athletes who have recognized the need for oral protection during their sporting activities use sports mouthguards as dental devices since many years [Knapik et al., 2007; Grippaudo et al. 2020]. According to the American Society for Testing and Materials three types of mouthguards exist: Type 1, stock mouthguards; Type 2, "boil and bite" or ready-made mouthguards; and Type 3, custom-made mouthguards. The most used mouthguards by athletes are those ready-made because of the low cost and easy access. Custom-made mouthguards are made using an impression of the individual's teeth and according to specifications provided by a dental professional. The material mainly used for making the mouthguard has become ethylene vinyl acetate (EVA) 0+ [ADADS, 2012]. Several studies of hockey and football players showed that "boil and bite" protective athletic mouthguards had the potential to become a microbial reservoir of pathogenic and opportunistic bacteria, yeasts, molds, and to increase the number and severity of oral mucosal injuries [Glass et al., 2009; Glass et al., 2011]. Custom-made mouthguards have many advantages compared to the stock and ready-made types: they show optimal comfort and good wearability [De Young et al., 2004; Spinas et al., 2014; Re et al., 2014], and they reveal no negative effects on elite taekwondo athletes' satisfaction. Moreover, they are protective device against orofacial injury and they allow for stable muscle activity during the training of Karate-Dō athletes [Bemelmanns et al, 2001; Eroğlu et al, 2006; Spinas et al., 2015; Raquel et al., 2016]. However, use of customised mouthguard in sports activities leads to changes in ecological factors of the oral cavity that inhibit the protective effect of saliva [D'Ercole at al., 2014; Mummolo et al., 2014 (b)].

For the evaluation of physiological biomarkers of sports training, the collection and analysis of saliva are rapidly developing as main evaluation tools. The collected saliva is used for monitoring quality, pH, buffering capacity, bacterial count, plaque, gingival inflammation and immune markers in sport and exercise. To date, saliva samples are an important part of a complete clinical check-up for many researchers [Papacosta and Nassis, 2011; Mummolo et al., 2014 (a); D'Ercole et al., 2016]. Saliva can provide a useful and non-invasive alternative to the collection of serum and plasma, because it can be collected rapidly, frequently and without stress. Furthermore, saliva collection requires less medical training and it can be performed on the sports field [D'Ercole et al., 2013; D'Ercole and Tripodi, 2013]. Isoprostanes, produced in membrane lipids in all body tissues through the

action of phospholipases, are generated by the free radical-induced and COX-independent peroxidation of essential polyunsatured fatty acids including arachidonic acid [Karamouzis et al., 2004]. Isoprostanes represent stable markers of oxidative stress *in vivo* and it has been measured in numerous tissues and body fluids including plasma, urine, exhaled breath condensate, saliva, cerebral spinal fluid and amniotic fluid [Milne et al., 2015]. Amongst F-2isoprostanes, 8-iso-prostaglandin F2α represents a stable biomarker of lipoperoxidation *in vivo* [Praticò, 2002].

Oxidative stress reflects an imbalance between the production of reactive oxygen species (ROS) and a suitable antioxidant action. The relationship between exercise and oxidative stress is very complex and depends on type, duration and intensity of training. Regular moderate training appears beneficial for oxidative stress and health; conversely, an acute and strenuous effort leads to an overproduction of ROS [Pingitore et al., 2005]. Prostaglandin E2, recognised as a biologically active factor in the 1960s, is considered a stable marker of inflammation, since it promotes local vasodilation, local attraction and activation of neutrophils, macrophages and mast cells in the early stages of inflammation [Kalinski, 2012; Petrini et al., 2012]. Thus, the purpose of the present study was to compare a group of individuals practicing competitive sports (basketball and volleyball) with a group of non-competitive individuals in a standardised clinical research. We determined the potential effects on salivary factors, such as pH value, volume, PGE2 and 8-iso-PGF2α levels by training and by use of customised mouthguards.

Materials and methods

Study population

The study population was composed of 80 subjects, all males: 40 athletes: 20 of them practice volleyball, 20 basketball (test group) and 40 subjects who attend a gym, at a non-competitive level (control group). As shown in Table 1, there was no difference between test and control group with respect to age and gender. There was difference between test and control group with respect to training amounts of time.

The basketball players attended the sport hall of Chieti (Italy) and the volleyball players attended the sport hall of Ortona (Chieti, Italy). The control group was recruited from consecutive first-time patients visiting the Department of Medical, Oral and Biotechnological Sciences, University of Chieti, in the period 2016–17.

The following inclusion criteria were considered.

1. Age: adults (age <18).
2. Absence of active carious lesion.
3. Dental treatment not in progress.
4. For the athletes, they require mouthguards.
5. For the controls, they were not practicing any competitive sports.

Patients were excluded from the study if they met any of the following exclusion criteria:

1. Periodontitis;
2. Partial or total removable prosthesis;
3. Poor medical conditions (diabetes, asthma), systemic antibiotics, or local antimicrobials in the 3 months preceding the study, patients under medication affecting the saliva flow rate.

During the study, a patient who started a medication affecting the saliva flow rate or the biofilm composition was immediately excluded. The selected subjects participated voluntarily in the study. Initially patients were given oral and written information on the purpose of the study. Written informed consent was signed by subjects (Privacy Law DL 196/2003). The collection and use of saliva was approved by the Ethics Committee of University "G. d'Annunzio", Chieti-Pescara, Italy.

A self-administered questionnaire was used to obtain data concerning a complete medical history, episodes or conditions affecting hard and soft tissues of the oral cavity, oral hygiene practices.

Clinical monitoring

The players (test group) were analyzed at baseline (T0), pre-training (T1), post-training with custom-made Ethylene-Vinyl-Acetate (EVA) mouthguards (T2), post-training without mouthguards (T3). The control group was analyzed only at baseline (T0). At T0, a clinical monitoring was performed and on each patient were recorded the number of decayed (D), missing (M) and filled (F) teeth (T) (DMFT) to assess caries prevalence according to WHO criteria [Allgrove et al., 2014]. To evaluate oral hygiene and periodontal status, the Plaque Index (PL+) according to Silness and Löe, and the Löe & Silness Gingival Bleeding (BOP+) were used, respectively. In addition, an oral examination of intraoral mucosa was performed and assessed the presence/absence of bad habits and/or parafunctional habits.

The patients avoided eating or drinking and they didn't brush their teeth at least 2 hours before taking the samples at all stages.

Saliva collection

On each athlete, in the 4 time points, and on the control group at T0, we stimulated saliva for determining PGE2 and 8-iso-PGF2α levels and it was collected with Salivette® (Sarstedt AG & Co., Germany). Moreover, patients chewed for 1 minute the cotton roll present in Salivette®. The cotton roll was placed into the test tube; then they were centrifuged according to the indication of the manufacturer. The saliva, deposited on the bottom of the test tube, was subjected to laboratory analysis. Saliva sample, for determining pH value and volume/ml, was collected with paraffin-chewing stimulation for 5 minutes. Saliva pH was calculated with a pH meter (Elettrofor XS instruments, Borsea, Italy). All procedures were done by one calibrated researcher.

Mouthguard manufacturing

Custom mouthguards were made in a dental laboratory on models cast from impressions and using EVA materials. Preliminary correction and adjustment in static and dynamic occlusion were performed for each newly fabricated mouthguard. Finally, each player was instructed on oral and mouthguard hygiene. Each subject repeated the indications received in oeder to correct some inaccuracies.

Radioimmunoassay (RIA)

Saliva samples were collected and PGE2 and 8-iso-PGF2α levels (pg/ml) were measured by radioimmunoassay (RIA), as previously reported [Chiavaroli et al., 2010; Menghini et al., 2016]. Briefly, specific anti-8-iso-PGF2α and anti-PGE2 were developed in the rabbit; the cross-reactivity against other prostanoids was <0.3%. Two hundred and fifty microliters of prostaglandin standard or sample were incubated overnight

	Total number	Male	Mean age	Training table/week
Basketball players	20	20	13,95 ± 2,54	1,93±0,18 h 5 times a week
Volleyball players	20	20	14,67 ± 2,22	2,06±0,53 h 5 times a week
Control group	40	40	13,13 ± 3,09	1,07±0,82 h 3 times a week

TABLE 1 Demographic characteristics of the studied population.

	Basketball players	Volleyball players	Control group
Dmft	5,13 ±3,2705	6,60±5,9833	4,55±3,0029
Daily Brushing Frequency	2,58±0,5339	3,11±0,5465	2,11±0,8965
PII+	64,71%	66,67%	80,00%
Previous Dental Trauma	30,00%	33,33%	13,33%
Mouthguard	15,00%	0,00%	7,14%
Dental Erosions	26,32%	22,22%	20,00%
Dental Stains	50,00%	55,56%	78,57%
Tooth Wear	56,25%	57,14%	66,67%
Recessions	47,06%	55,56%	40,00%

TABLE 2 Clinical parameters and prevalence (%) of clinical characteristics between genders at baseline.

at 4°C with the 3H-prostaglandin (3000 cpm/tube; NEN) and antibody (final dilution: 1:120 000), in a volume of 1.5 mL of 0.025 M phosphate buffer. Free and antibody-bound prostaglandins were separated by the addition of 100 µL 5% bovine serum albumin and 100 µL 3% charcoal suspension, followed by centrifugation for 10 min at 4,000 ×g at 5°C and decanting of the supernatants into the scintillation fluid (UltimaGold™, Perkin Elmer) for β emission counting. The detection limit of the assay method is 0.6 pg/mL.

Statistical analysis

Statistical analysis was performed using GraphPad Prism version 5.01 for Windows (GraphPad Software, San Diego, CA). Means ± S.E.M. were determined for each experimental group and analyzed by one-way analysis of variance (ANOVA), followed by Bonferroni post-hoc test. Statistical significance was accepted at $P < 0.05$.

Results

The demographic and clinical characteristics of the studied populations are presented in Table 1 and Table 2. Age is similar in the two groups. There was no difference with respect to amounts of time between the players.

As shown in Table 2 the analysed athletes have a very poor oral health, as demonstrated by high values of DMFT, PIL+, dental stains, tooth wear and gingival recessions, even if they brushed their teeth two or more times a day. Over 30% of elite players had suffered dental trauma related to sports, but only the 15% of basketball players used boil & bite mouthguard.

Our measurements showed an inhibition of 8-iso-PGF2α salivary release induced by physical exercise in each group of athletes compared to the control group (T0-T3). Moreover, there was a significant reduction of isoprostane level in T2 group compared to T0. In contrast, there was a significant increase in the salivary level of 8-iso-PGF2α in athletes who did not use the mouthguard with respect to groups at T1 and T2 (Fig. 1).

The saliva values of Prostaglandin E2 are reported in Figure 2. Compared to controls we had a significant increase in PGE2 in groups at T1 and T2, while in other conditions there were no significant changes on ProstaglandinE2 release. The observed increase at T2 was statistically significant compared to other time points groups. Additionally, we did not observe any difference in the collected saliva pH in control group and at T0-T3 stages (Fig. 3).

Finally, in saliva volume test produced in 5 minutes, a significant inhibition on saliva production emerged in T1-T3

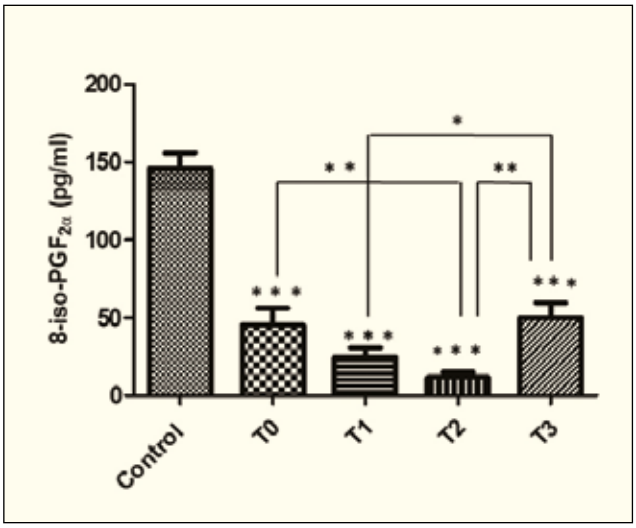

FIG. 1 8-iso-PGF2α production from saliva samples in the tested 4 time points.
T0: baseline.
T1: pre-training.
T2: post-training with custom-made EVA mouthguards.
T3: post-training without mouthguards.
Values represent the means ± SEM [ANOVA, $P < 0.0001$; post-hoc, *** $P < 0.001$ vs. Control (Control group), ** $P < 0.01$ vs. T2, ** $P < 0.01$ vs. T3, * $P < 0.05$ vs. T3].

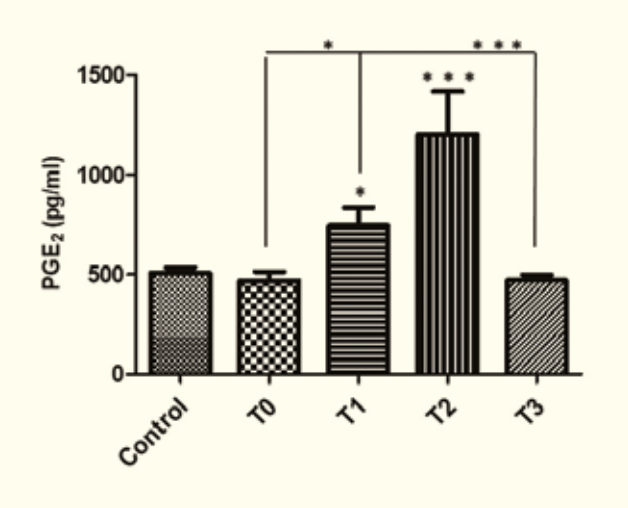

FIG. 2 PGE2 production from saliva samples in the tested 4 time points. Values represent the means ± SEM [ANOVA, p<0.0001; post-hoc, *** P< 0.001 vs. Control (Control group), * P < 0.05 vs. T1, * P < 0.05 vs. T0, * P < 0.05 vs. T1, *** P< 0.001 vs. T3].

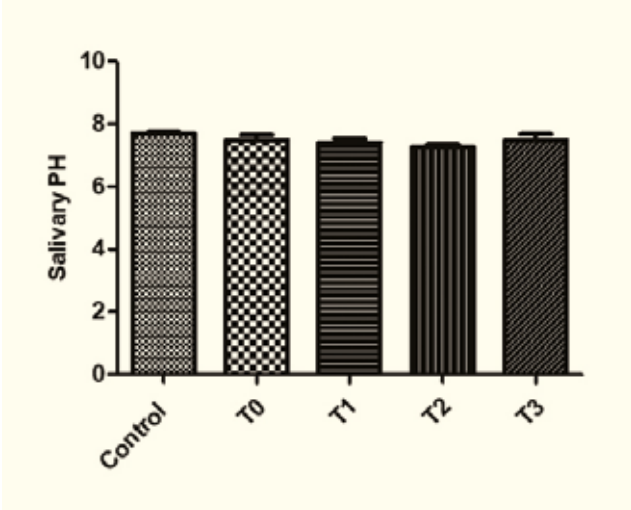

FIG. 3 Salivary pH from saliva samples in the tested 4 time points.

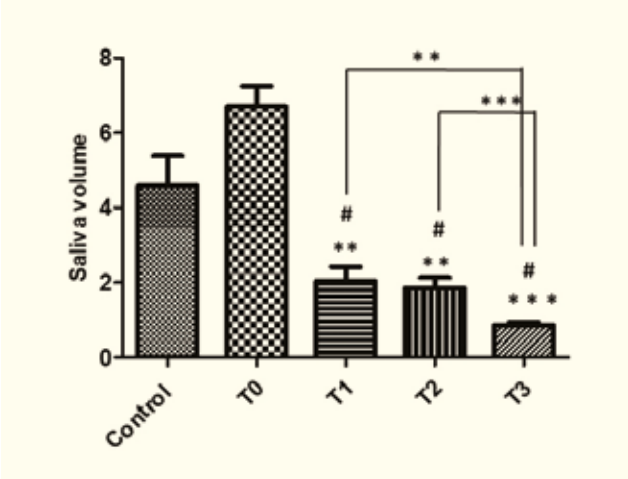

FIG. 4 Salivary volume in the tested groups. Values represent the means ± SEM [ANOVA, P <0.0001; post-hoc, *** P < 0.001 vs. Control (Control group), ** P < 0.01 vs. Control (Control group), #: *** P < 0.001 vs. T0, ** P < 0.01 vs. T3, *** P < 0.001 vs. T3].

stages with respect to the control group and to T0. A further significant reduction in saliva production also turned out at T3 compared to measurement at T1 and T2 (Fig. 4).

Discussion

High performance standards required of competitive players can only be attained by totally healthy individuals. Excellent dental care to players is a complicated task, due to numerous competitions that do not allow adequate planning of medical decisions. The athletes analyzed in this study have showed a very poor oral health, as demonstrated by high values of clinical index, as DMFT, PIL+. They also showed several oral conditions, such as dental stains, tooth wear and recessions. Despite, over 30% of elite players had suffered dental trauma related to sports, only the 15% of basketball players used boil & bite mouthguard.

The aim of this study was above all to raise awareness among young players for the use of custom-made mouthguards and the advantages deriving from their use. Normally instrumental tests, as electromyography (EMG) are used to evaluate to muscle fatigue, and/or muscle activity in different sports [Raquel et al., 2016; Quinzi et al., 2019; Tripodi et al., 2019]. Considering the athletes' young age, as an alternative to instrumental tests, in this study the comfort offered by custom-made mouthguards has been evaluated through the determination of effects on salivary factors, such as pH value, volume, levels of PGE2 and 8-iso-PGF2α, taking advantage of easy saliva sampling. As effect of training, a significant exercise-dependent shift of parameters revealed a decrease in saliva volume produced in 5 minutes, in athletes who didn ot wear the mouthguard during training. Conversely, mouthguard use does not significantly reduce saliva production. It is determined that physical exercise elicits increased sympathetic activation for which the reasons for the significant decrease from before to after training without mouthguard are due to an increase in sympathetic activity and a repression of the cholinergic parasympathetic innervation causing a significant vasoconstriction in salivary glands [Frese et al., 2014].

The volume of saliva is further altered also due to the deficit of liquids and electrolytes following the loss of water and sweat during physical exertion. These alterations in saliva flow rates are in line with previous investigations [Ferrazzano et al., 2010; Mulic et al., 2012; Tripodi et al., 2012; Allgrove et al., 2014]. On the contrary, the presence of a foreign body as a mouthguard works as a stimulating effect on the production of saliva. The mouthguard acts on the parasympathetic system. Parasympathetic cholinergic nerve stimulation seems to induce vasodilation of the capillaries supplying the salivary glands, thereby increasing blood flow; hence, elevated blood flow to the glands is associated with a higher saliva secretion rate [Papacosta and Nassis, 2011; Giuca et al., 2012]. The training and use of the mouthguard do not influence the pH of saliva collected in the various phases for both athletes and controls.

Normally, the oral cavity has the ability to adapt to the

presence of a foreign body, to alter the composition of saliva, to maintain high pH values and buffer power, and in this way prevents the colonization of potentially pathogenic microorganisms and maintain optimal environmental conditions. The salivary pH increased or decreased dependent on the beverages consumed by the athletes, but these aspects were not investigated in this study, and we would analyze them in further projects.

The isoprostanes are a family of eicosanoids of non-enzymatic origin produced by the random oxidation of tissue phospholipids by oxygen radicals. At least one of isoprostanes, 8-iso-PGF2α has been shown to have biological activity and it has been suggested as a marker of antioxidant deficiency, and lipid peroxidation [Karamouzis et al., 2004]. Particularly, lipid peroxidation has long been involved in tissue damage [Achitei et al., 2013]. Different researchers observed physical activity reduces 8-iso-PGF2α and it effectively attenuates systemic oxidative stress with considerable benefits for athletes [Haxhi et al., 2016; Lessiani et al., 2016]. Additionally, several studies revealed a lower oxidative stress status in trained athletes than in sedentary individuals [Bloomer et al., 2008; Falone et al., 2010].

Our data are in accordance with studies that revealed a decrease in oxidative stress for subjects who practice physical activity constantly. In this study, we observed a significant difference for the same athletes when they used the mouthguard or not during the game.

For athletes who did not wear the mouthguard during the training there was a significant decrease in 8-iso-PGF2α level compared to controls, even if the greatest reduction emerged in those who used the mouthguard. Recent studies showed that the use of the mouthguard improved physical performance by increasing in both aerobic and anaerobic activities the respiratory capacity of athletes [Schultz Martins et al., 2018]. In subjects with reduced respiratory capacity, as in patients with cystic fibrosis, the 8-iso-prostaglandin2α concentration was significantly higher than in healthy controls [Spicuzza et al., 2018].

Based on the results obtained we could hypothesise that the use of the mouthguard increases aerobic performance and leads to a further decrease in 8-iso-PGF2α release.

PGE2 is the main prostaglandin produced by cyclooxygenase (COX)-2 during inflammation and oxidative stress [Brunetti et al., 2012]. It has been known for many years that plasma concentrations of PGE2 increase during exercise and a high quantity may persist for up to two days [Pedersen et al., 1990; Smith et al.,1993]. Standardised physical exercise is associated with cyclo-oxygenase dependent formation of inflammatory prostaglandins. Intriguingly, we observed a significant increase in saliva PGE2 level at T2 in athletes who used the mouthguard. This increase is greater than enhanced production of PGE2 that we noticed in the same athletes before training or not using the mouthguard. In this context, further increase in prostaglandin E2 production could be related to use of the mouthguard. Some authors suggested that sport mouthguards can become a source of microbes, promote the development of oral and systemic diseases and alter oral environmental factors, inhibiting the protective effect of saliva [D'Ercole et al., 2017; D'Ercole et al., 2020]. In several studies, an increase in salivary PGE2 has been observed in the transitional phase between health and periodontal disease [Syndergaard et al., 2014; Wang et al., 2017]. On the other hand, we cannot exclude that PGE2 increase at T2 in athletes could be the result of a prostaglandin-induced pleiotropic effect. PGE2 has been reported to stimulate muscle relaxation [Insuela et al., 2015], while the use of mouthguard could improve the activity of masticatory muscles in Karate-Dō athletes [Raquel et al., 2016].

Furthermore, we could substantiate the onset of protective effects induced by mouthguard use in athletes. Particularly, the stimulated PGE2 levels could indicate a possible improvement of masticatory muscle function thus leading to a minor risk of tissue damage, as revealed by reduced level of 8-iso-PGF2α.

Conclusions

As a diagnostic tool for the assessment of training conditions we can use both the levels of salivary and plasmatic steroid hormones observed during exercise.

To prevent dental trauma, the screening of caries, periodontal disease and other oral pathologies in the young athletes by means of oral examinations before the beginning of the sport season and periodical follow-ups by dentists are very important. Until now, preventive and periodic examinations are not carried out even in high-level professional sports teams, such as in the case of football.

Our results suggest a possible association between sports activity and reduction in oxidative stress especially in athletes who use the mouthguard.

Finally, even if further investigations will be necessary with a greater number of athletes engaged in different competitive disciplines the use of mouthguards could be useful for athletes because it could increase their respiratory capacity and their muscle functions and protect them from orofacial injuries.

Author Contributions

Conceptualization, S.D. and D.T.; methodology, C.F. and L.R..; software, D.F.; validation, C.F. and L.R.; formal analysis, A.C.; investigation, S.D. and D.T.; resources, C.F.; data curation, D.F.; writing—original draft preparation, A.C. and S.D.; writing—review and editing, S. D. and A.C.; visualization, L.R.; supervision, S.D. and D.F.; project administration, D.T.; funding acquisition, S.D. and D.T.

Acknowledgments

This work was supported by the Department of Medical, Oral and Biotechnological Sciences, "G. d'Annunzio" University of Chieti, Italy.

Conflicts of Interest

Authors declare that they have no conflict of interest.

References

› Achitei D, Ciobica A, Balan G, Gologan E, Stanciu C, Stefanescu G. Different profile of peripheral antioxidant enzymes and lipid peroxidation in active and non-active inflammatory bowel disease patients. Dig Dis Sci 2013; 58:1244-9.
› Allgrove JE, Oliveira M, Gleeson M. Stimulating whole saliva affects the response of antimicrobial proteins to exercise. Scand J Med Sci Sports 2014; 24: 649.
› American Dental Association Division of Science. For the dental patient: Keep sports safe–wear a mouthguard. JADA 2012; 143: 312.
› Bemelmanns, P, Pfeiffer P. Shock absorption capacities of mouthguards in different types and thicknesses. Int J Sports Med 2001; 22: 149-53.
› Bloomer RJ, Fisher-Wellman KH. Blood oxidative stress biomarkers: influence of sex, exercise training status, and dietary intake. Gend Med

2008; 5: 218–28.
› Brunetti L, Leone S, Chiavaroli A, Orlando G, Recinella L, Ferrante C, Di Nisio C, Verratti V, Vacca M. Cafeteria diet increases Prostaglandin E2 levels in rat prostate, kidney and testis. Int J Immunopathol Pharmacol 2012; 23: 1073-8.
› Chiavaroli A, Brunetti L, Orlando G, Recinella L, Ferrante C, Leone S, Di Michele P, Di Nisio C, Vacca M. Resveratrol inhibits isoprostane production in young and aged rat brain. J Biol Regul Homeost Agents 2010; 24: 441–446.
› D'Ercole S, Martinelli D, Tripodi D. Influence of sport mouthguards on the ecological factors of the children oral cavity. BMC Oral Health 2014; 14: 97.
› D'Ercole S, Ristoldo F, Quaranta F, Amaddeo P, Tripodi D. Analysis of oral health status and of salivary factors in young soccer players: a pilot study. Medicina dello sport 2013; 66: 71-80.
› D'Ercole, S, Tripodi, D. The effect of swimming on oral ecological factors. J Biol Regul Homeost Agents 2013; 27: 551-8.
› D'Ercole S, Tieri M, Martinelli D, Ciaravino C, Fulco D, Tripodi D. Microbial Contamination and Disinfection of Sport Mouthguard: In Vitro Study. Curr Microbiol. 2020; 77: 246-253.
› D'Ercole S, Tieri M, Martinelli D, Tripodi D. The effect of swimming on oral health status: competitive versus non-competitive athletes. J Appl Oral Sci 2016; 24: 107-13.
› D'Ercole S, Tieri M, Martinelli D, Tripodi D. The use of chlorhexidine in mouthguards. J Biol Regul Homeost Agents 2017; 31:487-493.
› DeYoung AK, Robinson E, Godwin WC. Comparing comfort and wearability: custom-made vs. self-adapted mouthguards. J Am Dent Assoc 2004; 125: 1112-1118.
› Eroğlu, E, Diljin, KA, Lu¨tfi BM. Elite taekwondo athletes' satisfaction with custom-made mouthguards. Dent Traumatol 2006, 22: 193-197.
› Falone S, Mirabilio A, Pennelli A, Cacchio M, Di Baldassarre A, Gallina S, Passerini A, Amicarelli F. Differential impact of acute bout of exercise on redox- and oxidative damage-related profiles between untrained subjects and amateur runners. Physiol Res 2010; 59: 953–61.
› Ferrazzano GF, Orlando S, Ingenito A, Tia M, Sammartino G. Tooth replantation as an alternative to dental implantology in adolescent patients. Eur J Paediatr Dent 2010; 11: 216-218.
› Frese C, Frese F, Kuhlmann S, Saure D, Reljic D, Staehle HJ, Wolff D. Effect of endurance training on dental erosion, caries, and saliva. Scand J Med Sci Sports 2014; 25: 319-326.
› Giuca MR, Pasini M, Nastasio S, D' Ercole S, Tripodi D. Dental and periodontal complications of labial and tongue piercing. J Biol Regul Homeost Agents. 2012; 26: 553-60.
› Glass RT, Conrad RS, Köhler GA, Warren AJ, Bullard JW. Microbiota found in protective athletic mouthguards. Sports Health 2011; 3, 244–248.
› Glass RT, Wood CR, Bullard JW, Conrad RS. Protective Athletic Mouth Guards: Do They Cause Harm? Sports Health 2009; 1: 411-5.
› Grippaudo C, Quinzi V, Manai A, Paolantonio EG, Valente F, La Torre G, Marzo G. Orthodontic treatment need and timing: Assessment of evolutive malocclusion conditions and associated risk factors. Eur J Paediatr Dent 2020; 21: 203-8.
› Haxhi J, Leto G, di Palumbo AS, Sbriccoli P, Guidetti L, Fantini C, Buzzetti R, Caporossi D, Di Luigi L, Sacchetti M. Exercise at lunchtime: effect on glycemic control and oxidative stress in middle-aged men with type 2 diabetes. Eur J Appl Physiol 2016; 116: 573-82.
› Insuela DB, Daleprane, JB, Coelho LP, Silva AR, Silva PM, Martins MA, Carvalho VF. Glucagon induces airway smooth muscle relaxation by nitric oxide and prostaglandin E_2. J Endocrinol 2015; 225: 205-17.
› Kalinski, P. Regulation of immune responses by prostaglandin E2. J Immunol. 2012; 188: 21-28.
› Karamouzis I, Christoulas K, Grekas D, Giannoulis K, Vamvakoudis E, Mandoukas K. The response of muscle interstitial F2-isoprostane (8-ISO-PGF2alpha) during dynamic muscle contractions in humans. Prostaglandins Leukot Essent Fatty Acids 2004; 71: 87-90.
› Knapik, JJ, Marshall, SW, Lee RB, Darakjy SS, Jones SB, Mitchener TA, DelaCruz GG, Jones BH. Mouthguards in sport activities: history, physical properties and injury prevention effectiveness. Sports Med 2007; 37: 117–144.
› Lessiani G, Santilli F, Boccatonda A, Iodice P, Liani R, Tripaldi R, Saggini R, Davì G. Arterial stiffness and sedentary lifestyle: Role of oxidative stress. Vascul Pharmacol 2016; 79: 1-5.
› Menghini, L, Ferrante C, Leporini L, Recinella L, Chiavaroli A, Leone S, Pintore G, Vacca M, Orlando G, Brunetti L. An hydroalcoholic chamomile extract modulates inflammatory and immune response in HT29 cells and isolated rat colon. Phytotherapy research 2016; 30: 1513-1518.
› Milne GL, Dai Q, Roberts Ii LJ. The isoprostanes—25 years later. Biochim. Biophys. Acta Mol. Cell Biol. Lipids 2015; 1851: 433–445.
› Mulic A, Tveit AB, Songe D, Sivertsen H, Skaare AB. Dental erosive wear and salivary flow rate in physically active young adults. BMC Oral Health 2012; 12: 8.
› Mummolo S, D'Ercole S, Marchetti E, Campanella V, Martinelli D, Marzo G, Tripodi D. Oral antiseptic and periodontitis: a clinical and microbiological study. Oral Health Dent Manag. 2014; 13: 698-702. (a)
› Mummolo S, Tieri M, Tecco S, Mattei A, Albani F, Giuca MR, Marzo G. Clinical evaluation of salivary indices and levels of Streptococcus mutans and Lactobacillus in patients treated with Occlus-o-Guide. Eur J Paediatr Dent. 2014; 15: 367-70.(b)
› Papacosta E, Nassis GP. Saliva as a tool for monitoring steroid, peptide and immune markers in sport and exercise science. J Sci Med Sport 2011; 14: 424-34.
› Pedersen BK, Tvede N, Klarlund K, Christensen LD, Hansen FR, Galbo H, Kharazmi A, Halkjaer-Kristensen J. Indomethacin in vitro and in vivo abolishes post-exercise suppression of natural killer cell activity in peripheral blood. Int J Sports Med 1990; 11: 127-31.
› Petrini M, Ferrante M, Ciavarelli L, Brunetti L, Vacca M, Spoto G. Prostaglandin E2 to diagnose between reversible and irreversible pulpitis. Int. J. Immunopathol. Pharmacol. 2012; 25: 157–163.
› Pingitore A, Lima GP, Mastorci F, Quinones A, Iervasi G, Vassalle C. Exercise and oxidative stress: potential effects of antioxidant dietary strategies in sports. Nutrition 2005; 31: 916-922.
› Praticò, D. Alzheimer's disease and oxygen radicals: new insights. Biochem Pharmacol 2002; 63: 563-567.
› Quinzi V, Rossi O, Paglia L, Marzo G, Caprioglio A. Paediatric Orthodontics Part 2: Periodontal effects of maxillary expansion on teeth in deciduous and mixed dentition. Eur J Paediatr Dent 2019; 20: 164-166.
› Raquel G, Namba EL, Bonotto D, Ribeiro Rosa EA, Trevilatto PC, Naval Machado MÂ, Vianna-Lara MS, Azevedo-Alanis LR. The use of a custom-made mouthguard stabilizes the electromyographic activity of the masticatory muscles among Karate-Dō athletes. J Bodyw Mov Ther 2016; 21: 109-116.
› Re D, Augusti D, Paglia G, Augusti G, Cotti E. Treatment of traumatic dental injuries: evaluation of knowledge among Italian dentists. Eur J Paediatr Dent 2014; 15: 23-8.
› Schultz Martins R., Girouard P, Elliott E, Mekary S. Physiological Responses of a Jaw-Repositioning Custom-Made Mouthguard on Airway and Their Effects on Athletic Performance. J Strength Cond Res 2018; doi: 10.1519/JSC.0000000000002679.
› Smith LL, Wells JM, Houmard JA, Smith, S.T, Israel RG, Chenier, TC, Pennington SN. Increases in plasma prostaglandin E2 after eccentric exercise. A preliminary report. Horm Metab Res 1993; 25: 451-2.
› Spicuzza L, Parisi GF, Tardino L, Ciancio N, Nenna R, Midulla F, Leonardi S. Exhaled markers of antioxidant activity and oxidative stress in stable cystic fibrosis patients with moderate lung disease. J Breath Res, 2018; 12(2):026010.
› Spinas E, Aresu M, Giannetti L. Use of mouth guard in basketball: observational study of a group of teenagers with and without motivational reinforcement. Eur J Paediatr Dent 2014; 15: 392-6.
› Spinas E, Aresu M, Canargiu F, Giannetti L. Preventive treatment of post-traumatic dental infraocclusion: study on the knowledge of dental decoronation in a sample of Italian dental students and dentists. Eur J Paediatr Dent 2015; 16: 279-283
› Spinas E, Giannetti L, Mameli A, Re D. Dental injuries in young athletes, a five-year follow-up study. Eur J Paediatr Dent 2018; 19: 187-193
› Syndergaard B, Al-Sabbagh M, Kryscio RJ, Xi J, Ding X, Ebersole JL, Miller CS. Salivary biomarkers associated with gingivitis and response to therapy. J Periodontal 2014; 85: 295-303.
› Tripodi, D, Fulco D, Beraldi A, Ripari P, Izzi G, D'Ercole S. Custom-Made Mouthguards: Electromyographic Analysis of Masticatory Muscles and Cardiopulmonary Tests in Athletes of Different Sports. Health 2019; 11: 428-438.
› Tripodi D, Perrotti V, Latrofa M, D'Ercole S, Artese L, Piattelli A. Management of compound odontoma in a 10-year-old girl preserving the associated impacted permanent tooth. Eur J Paediatric Dent 2012; 13: 268-271.
› Wang Y, Ren B, Zhou X, Liu S, Zhou Y, Li B, Jiang Y, Li Feng M, Cheng L. Growth and adherence of Staphylococcus aureus were enhanced through the PGE2 produced by the activated COX-2/PGE2 pathway of infected oral epithelial cells. PLoS One 4 2017; 12: e0177166.